

1 **The teat cup detachment level affects milking performance in an automatic milking**  
2 **system with teat cleaning and milking in the same teat cup**

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**Supplementary File**

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## 13 **Material & Methods**

### 14 *Animals and Housing*

15 The cows were divided into two groups according to their lactational stage. The distribution of  
16 the lactation numbers were similar in the two groups with one having an average of lactation  
17 number 3.17 (SEM  $\pm$  1.11) and the other 3.83 (SEM  $\pm$  1.37).

18 The cows were housed in a free-stall barn with directed cow traffic. Finger gates were installed  
19 between the feeding and lying area to prevent cows from going to the feeding area without  
20 passing through a sorting gate. The sorting gate was installed in front of the waiting area for  
21 the automatic milking system (AMS). Sorting was based on the minimal milking interval set  
22 by the farmer for each cow. If a cow did not yet reach the minimal milking interval when  
23 entering the sorting gate, it did not have milking permission and was led through a corridor to  
24 the feeding area. Cows with milking permission were led into the waiting area which could  
25 only be left by passing through the AMS. Minimal milking intervals were chosen mostly  
26 considering the milk yield per milking. High yielding cows were milked more frequently.  
27 Other factors affecting the decision amongst other things were udder health, frequency in  
28 passing the sorting gate and teat position where udder filling affected the ability of the AMS to  
29 attach the teat cups.

### 30 *Milking*

31 Milking in the DairyRobot R9500 (GEA, Bönen, Germany) works differently than most AMS  
32 systems used for scientific studies. The sequence of processes in the standard milking routine  
33 is pictured in **Figure S1**. After the cow enters the AMS, the milking cluster moves from its side  
34 compartment to position itself under the cow. As soon as it's in position under the udder the  
35 stimulation time starts running, which is set for the whole milking cluster. The stimulation time  
36 is set depending on the DIM and can be adjusted by the farmer. The settings that were used on  
37 the farm are shown in **Table S1**. The AMS used StimoPuls technology (GEA, Bönen,  
38 Germany) which reduces the pulsation chamber vacuum (PCV) to 20 kPa to keep the liner  
39 closed and vibrates with a pulsation rate of 300 pulses per minute. When the pre-set stimulation  
40 time is reached, PCV and pulsation switch to the regular milking settings (41 kPa, 64  
41 pulses/min) in all teat cups at once, and milking starts.

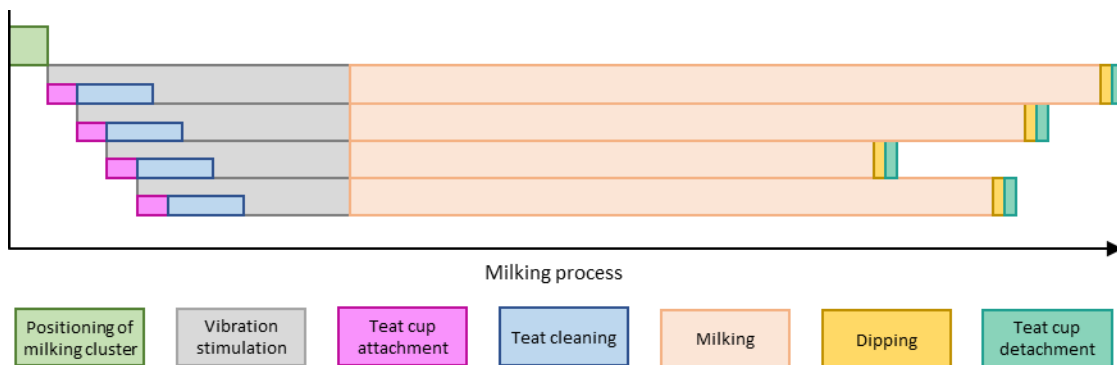
42 The attachment of the teat cups starts with the hind teats but does not follow a specific order.  
43 The teats are labelled to their respective teat cups by the G5 Time-of-Flight camera. A time-  
44 of-flight camera is a camera system that uses infrared light to determine distance between the

45 camera and the subject for each point of the image, by measuring the round trip time of the  
 46 light. Whether the left or right teat is attached first depends on which teat is first detected by  
 47 the camera. Teats are attached one after the other and not simultaneously. Attachment took on  
 48 average 6 seconds per quarter. Teat cleaning starts as soon as the sensor registers the teat  
 49 attachment. The cleaning duration was set to 10 seconds.

50 Milking starts after the stimulation time runs out. If a teat cup has not been attached by the time  
 51 the stimulation time runs out, the teat will be cleaned and milked directly afterwards without  
 52 any stimulation on this teat. Because the teats are mostly attached and cleaned before the  
 53 stimulation time ends, milking usually starts in all quarters together. If a teat takes longer than  
 54 the stimulation time to attach, it will be washed while the other teats are being milked. The  
 55 milking in the remaining quarter will start when teat cleaning is finished.

56 Vacuum and pulsation are switched off after the milk flow drops below the pre-set quarter milk  
 57 flow threshold for detachment for 5 seconds. Dipping agent is applied to the teat with a spray  
 58 function within the teat cup before the teat cup is removed.

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62 **Supplementary Figure S1.** Sequence of processes in the standard milking routine

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65 **Supplementary Table S1.** Stimulation settings on the farm

Days in milk	1	20	150	200	365	600
Stimulation (sec)	40	50	60	75	75	90

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67 **Supplementary Table S2.** Means and standard error of milking time dependent of detachment at below and above average milking interval in  
68 early and late lactating cows.

<b>Lactation stage</b>		<b>early lactation</b>			<b>late lactation</b>		
<b>Detachment [g/min]</b>		<b>50</b>	<b>87.5</b>	<b>125</b>	<b>50</b>	<b>87.5</b>	<b>125</b>
	Milking interval						
<b>hind right</b>	<10h	5.75 ± 0.21 <sup>bα</sup>	4.63 ± 0.20 <sup>bβ</sup>	4.60 ± 0.17 <sup>bβ</sup>	4.81 ± 0.23 <sup>α</sup>	4.32 ± 0.15 <sup>bβ</sup>	3.45 ± 0.29 <sup>β</sup>
	>10h	6.71 ± 0.27 <sup>αα</sup>	5.97 ± 0.23 <sup>aβ</sup>	5.26 ± 0.34 <sup>aβ</sup>	5.26 ± 0.31 <sup>α</sup>	4.57 ± 0.22 <sup>αα</sup>	4.41 ± 0.21 <sup>β</sup>
<b>hind left</b>	<10h	5.56 ± 0.27 <sup>α</sup>	4.34 ± 0.26 <sup>β</sup>	4.29 ± 0.20 <sup>bβ</sup>	4.99 ± 0.22 <sup>α</sup>	3.71 ± 0.18 <sup>bβ</sup>	3.49 ± 0.31 <sup>β</sup>
	>10h	6.21 ± 0.30	5.39 ± 0.24	4.90 ± 0.26 <sup>a</sup>	4.96 ± 0.33 <sup>α</sup>	4.48 ± 0.25 <sup>ααβ</sup>	4.18 ± 0.24 <sup>β</sup>
<b>front right</b>	<10h	5.22 ± 0.33 <sup>α</sup>	4.04 ± 0.23 <sup>bβ</sup>	4.32 ± 0.21 <sup>β</sup>	3.91 ± 0.20 <sup>α</sup>	3.18 ± 0.19 <sup>bβ</sup>	2.89 ± 0.17 <sup>bβ</sup>
	>10h	5.43 ± 0.26 <sup>α</sup>	5.17 ± 0.24 <sup>ααβ</sup>	4.03 ± 0.23 <sup>β</sup>	4.30 ± 0.25 <sup>α</sup>	3.68 ± 0.25 <sup>ααβ</sup>	3.40 ± 0.25 <sup>aβ</sup>
<b>front left</b>	<10h	5.42 ± 0.39 <sup>Aα</sup>	3.84 ± 0.24 <sup>bβ</sup>	4.08 ± 0.22 <sup>β</sup>	3.68 ± 0.27 <sup>Bα</sup>	3.36 ± 0.20 <sup>aβ</sup>	2.97 ± 0.18 <sup>bβ</sup>
	>10h	6.00 ± 0.24 <sup>Aα</sup>	5.33 ± 0.28 <sup>Aαβ</sup>	3.98 ± 0.22 <sup>β</sup>	3.97 ± 0.29 <sup>B</sup>	3.55 ± 0.17 <sup>B</sup>	3.41 ± 0.22 <sup>a</sup>

69 Lower-case letters: No common letter means a significant difference ( $P < 0.05$ ) between the milking intervals within a detachment level and  
70 lactation stage.

71 Upper-case letters: No common letter means a significant difference ( $P < 0.05$ ) between the lactation stage within a detachment level and  
72 milking interval.

73 Greek letters: No common letter means a significant difference ( $P < 0.05$ ) between the detachment settings within a milking interval and  
74 lactation stage.

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80 **Supplementary Table S3.** Means and standard error of teat tissue thickness dependent of detachment at below and above average milking  
 81 interval in early and late lactating cows.

<b>Lactation stage</b>		<b>early lactation</b>			<b>late lactation</b>		
<b>Detachment [g/min]</b>		<b>50</b>	<b>87.5</b>	<b>125</b>	<b>50</b>	<b>87.5</b>	<b>125</b>
<b>Left barrel</b>	Milking interval <10h	12.1 ± 0.3	11.6 ± 0.3	12.5 ± 0.3	11.3 ± 0.2	11.3 ± 0.2	11.1 ± 0.4
	>10h	12.0 ± 0.3	12.7 ± 0.3	11.5 ± 0.3	11.4 ± 0.3	11.7 ± 0.2	11.2 ± 0.2
<b>Left tip</b>	<10h	11.5 ± 0.2	11.4 ± 0.2	11.8 ± 0.2	11.0 ± 0.2	11.2 ± 0.2	11.0 ± 0.2
	>10h	11.2 ± 0.2	11.8 ± 0.2	11.1 ± 0.2	11.1 ± 0.2	11.0 ± 0.2	10.9 ± 0.2
<b>Right barrel</b>	<10h	11.9 ± 0.3	11.5 ± 0.3	12.5 ± 0.3	11.3 ± 0.2	11.0 ± 0.2	11.2 ± 0.3
	>10h	11.5 ± 0.3	11.8 ± 0.3	10.6 ± 0.3	11.2 ± 0.2	11.3 ± 0.2	10.9 ± 0.2
<b>Right tip</b>	<10h	11.5 ± 0.2	11.4 ± 0.2	11.9 ± 0.2	11.1 ± 0.2	11.2 ± 0.1	11.2 ± 0.3
	>10h	11.1 ± 0.3	11.6 ± 0.3	11.2 ± 0.2	11.3 ± 0.3	11.2 ± 0.2	11.1 ± 0.2